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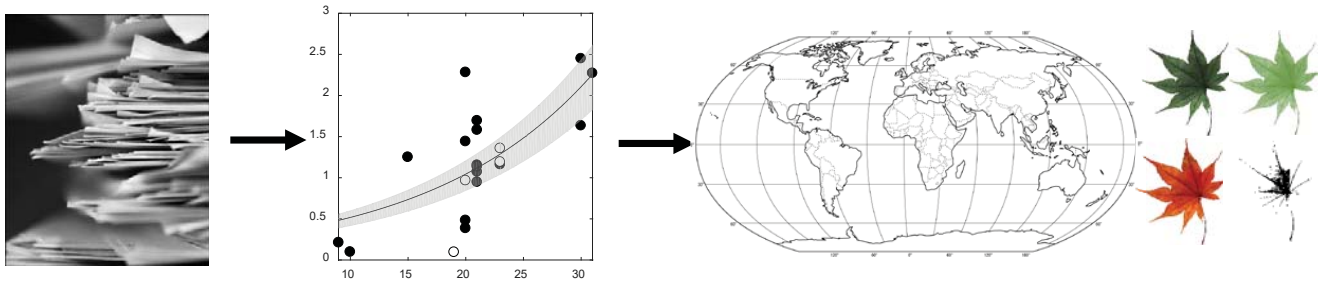
***Graphical Abstract**



Highlights:

- Transformation of illicit drug biomarkers in wastewater can be temperature-dependent.
- Relevant published scientific literature were systematically reviewed and selected for data collection.
- Arrhenius equation was used to describe temperature-dependent transformation kinetics of selected biomarkers in wastewater under aerobic conditions.
- The study can facilitate comparative assessments of drug stability in wastewater and more accurate estimation of drug consumption, especially in multi-catchment studies covering wide geographical area

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Abstract

Temperature is well-known as a key factor, influencing the transformation kinetics of organic chemicals. In the context of wastewater-based epidemiology; however, temperature differences among sewer catchments and within the same catchment (due, e.g., to seasonal variations) have been neglected to date as a factor influencing the estimation of illicit drug consumption.

In this study, we assessed the influence of temperature on the transformation of drug biomarkers occurring at trace levels in wastewater, based on laboratory-scale experimental evidence, and its ensuing implications for back-calculation of chemical consumption rate. Existing literature on the stability of drug biomarkers in untreated wastewater was systematically reviewed, and transformation rates obtained at different temperatures were collected. Arrhenius-based equations were fitted to empirical data and identified to describe the transformation of selected cocaine and morphine biomarkers at applicability temperature range (from 2–9ºC to 30–31ºC). These empirically-derived relationships were used to assess the influence of temperature on the transformation of drug biomarkers during in-sewer transport and its effect on the back-calculation of drug consumption rate in urban catchments. Findings from this study can help reduce the uncertainty intrinsic to wastewater-based epidemiology studies, and will be beneficial in generalizing consumption estimates from different catchments worldwide.

Keywords: Wastewater-based epidemiology, stability, temperature, biotransformation, Arrhenius equation

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1. Introduction

Wastewater-based epidemiology (WBE) is a growing research field to improve social behavior predictions in an epidemiological context. It is based on the analysis of substance residues (biomarkers) in wastewater and back-calculation of population consumption/exposure at catchment level. Substance use biomarkers, such as illicit drugs, have been the main focus of WBE studies (Gracia-Lor et al., 2017). A number of uncertainties (e.g., chemical analysis, determination of catchment population) have been associated to the determination of community drug use (Castiglioni et al., 2013). Furthermore, neglecting in-sewer transformation can also be a significant source of bias since biomarker concentration levels at the excretion point can differ from the sampling point (Li et al., 2018). In-sewer stability of drugs is mainly associated to abiotic transformation (without the presence of biomass) and biotransformation in the presence of suspended and attached biomass.

In WBE studies, two main approaches have been used to translate measured concentration to consumption rate: (i) lumped correction factors that include e.g., excretion ratios and in-sewer transformation; (ii) in-sewer process kinetic models together with excretion ratios. The first approach is commonly used due to its simplicity, with the major drawback of lacking catchment specificity. Conversely, process models explicitly rely on first- or second-order equations (McCall et al., 2016; Plósz et al., 2013; Ramin et al., 2016) to describe transformation kinetics, therefore allowing to account for a number of influencing factors (e.g. redox conditions, in-sewer residence time, transformation pathways and biomarker concentrations) depending on the complexity level. A factor known to influence microbial activity—hence biomarker stability—is temperature. The impact of temperature on the transformation of organic micropollutants has been assessed in activated sludge (Li et al., 2005) and in anaerobic digestion (Carballa et al., 2007). As to illicit drug biomarkers, stability studies in untreated wastewater (Bisceglia and Lippa, 2014b; Devault et al., 2017) have overall revealed enhanced

transformation kinetics with increasing temperature. While the effect of temperature on microbial growth kinetics is considered in models for conventional pollutants (e.g., activated sludge models), very few examples exist on quantifying the temperature dependence of kinetic model parameters for trace organic chemical transformation (Li et al., 2005; Wick et al., 2009).

In sewers, wastewater temperature exhibits seasonal and geographical variations and may further vary within the same catchment. During a recent Europe-wide sampling campaign (conducted simultaneously in 47 cities), the temperature of raw wastewater at sampling points was reported in the range between 7 ºC and 28ºC (Ort et al., 2014). Consequently, the impact of temperature on the stability of drug biomarkers in sewers may significantly vary from catchment to catchment, and the associated uncertainties propagating to the back-calculated consumption rate could be reduced in WBE approaches using more robust temperature models – the main focal area chosen for this study.

Considering existing limitations, the objectives of this study were: (i) to assess the effect of temperature on in-sewer drug biomarker stability, based on findings from published literature; (ii) Use empirical equations to describe temperature-dependent transformation kinetics of selected biomarkers in wastewater under aerobic conditions; (iii) to assess the influence of temperature on the in-sewer removal of drug biomarkers in a hypothetical urban catchment.

2. Materials and methods

2.1. Literature review and data treatment

Published scientific literature was reviewed (last update: 31/03/2018) to select drug biomarker stability studies in untreated wastewater, i.e. without the influence of biofilm. Further screening was performed to identify studies that fulfilled the following criteria: (i) stability studies were performed under aerobic conditions; (ii) biomarker transformation kinetics were explicitly reported or could be derived (calculated) based on presented results (e.g., concentration profiles in batch experiments); (iii) estimation of model parameters (see Eq. 1) was associated with good match between measured and predicted concentration profiles (R^2>0.7). Ten literature studies were eventually selected (Table 1), providing relevant information on stability of cocaine (COC), ecgonine methyl ester (EME), cocaethylene (CE), norcocaine (NorCOC) and 6-monoacetylmorphine (6-MAM).

The first-order transformation rate coefficient (k, d^{-1}) was used as indicator of biomarker stability in wastewater. Notably, k accounts for both abiotic and biotransformation kinetics, given that abiotic control experiments were absent in most of the selected studies. When k values were not explicitly reported, they were estimated by fitting experimental data with a first-order kinetic equation (Eq. 1):

$$C(t) = C_0 \, e^{-kt} \qquad \text{(Eq. 1)}$$

where C_0 and $C(t)$ are biomarker concentrations at time 0 and at time t, respectively.

In two cases (McCall et al., 2016; Ramin et al., 2016), abiotic and biotransformation kinetics were separately assessed and quantified by estimating the first-order rate coefficients (k_{abio}, d^{-1}) and pseudo-first-order rate coefficients (k_{bio}, L g^{-1} d^{-1}), respectively. The two kinetic indicators were combined to obtain k (Eq. 2):

$$k = k_{abio} + k_{bio} X_{TSS} \qquad \text{(Eq. 2)}$$

where X_{TSS} (g L^{-1}) denotes the concentration of total suspended solids (TSS) in the experiments. Data from concentration profiles were extracted, when necessary, using the software *PlotDigitizer*.

For each biomarker, the Arrhenius equation (Eq. 3) was used to describe variations in transformation rates as a function of temperature:

$$k_T = k_{25}\,\theta^{(T-25)} \qquad \text{(Eq. 3)}$$

where T(°C) denotes the temperature, at which a specific k_T value was derived, k_{25} the transformation rate at 25°C and θ (-) the exponential Arrhenius coefficient. Parameters θ and k_{25} were estimated for each biomarker using particle swarm optimization in MATLAB 2016b. A temperature of 25°C was selected as reference to improve the identifiability of both estimated parameters, as previously suggested (Schwaab et al., 2007).

<Table 1>

2.2. Back-calculation procedure

To back-calculate drug concentration at the release point e.g. after toilet flush (unknown), drug concentration at the influent of wastewater treatment plant (known) is considered in a hypothetical catchment. In-sewer transformation was simulated using Eq. 1 and assuming an average residence time of 4.5 h, corresponding to the average residence time in a recent European monitoring campaign (Ort et al., 2014). To reflect on the uncertainty of the estimated model parameters (e.g. k_T (d^{-1}) and Θ) Monte Carlo simulations with Latin hypercube sampling (LHS) were performed. To evaluate the impact of temperature on the removal of the selected drugs, three temperature conditions were considered, being representative of low (T=5°C), medium (T=15°C) and high (T=25°C) temperature.

3. Results and discussion

3.1. *Temperature-dependent transformation*

Considerable data variability in *k* rate values found in literature was noticed for most all selected drugs (Fig. 1), even considering the same temperature (e.g., 6-MAM) as a result of factors such as, differences in stability test conditions used in literature. Nevertheless, overall increase of *k* with increasing temperature was observed, especially when considering the mean of multiple measurements for each unique temperature.

Additionally, for each biomarker, Figure 1 presents plots of fitted Arrhenius equations (and associated 95% confidence intervals, shaded areas). Interestingly, many of the calculated data points (not reported in the original study) and estimated ones (reported in the original study) fall out of the confidence interval. Besides the previously discussed inherent data variability, this may have resulted from the limited applicability of first-order transformation kinetics e.g. due to significant microbial growth during batch experiments (Ramin et al., 2016).

Estimated parameter values k_T (d^{-1}) and Θ for the selected biomarkers are reported in Table 2. It can be noticed that the estimated relative error was low, below 50%, except for NorCOC (0.78%) and parameter collinearity was low expect for EME (-0.75). This seems to suggest good parameter identifiability, based on criteria (error < 50% and collinearity < 0.7) set by (Frutiger et al., 2016). Nevertheless, these thresholds are subjective and the consideration of 25°C as reference temperature allowed for the improvement of parameter identifiability (achieving lower correlation).

Estimated Θ coefficient values were between 1.04 and 1.18, in agreement with previously reported values. That is, for primary metabolic processes (relevant for biomass growth) in sewers, Arrhenius-based temperature corrections have been suggested, with Θ values of 1.07 and 1.05 for aerobic water

phase and biofilm processes, respectively (Hvitved-Jacobsen et al., 2013). Henze et al. (2000) also suggested similar coefficients to describe temperature dependency of biological processes in the Activated sludge model No. 2 (ASM2). These coefficients are ranging from low (Θ = 1.04) for hydrolysis by phosphate-accumulating biomass to high (Θ = 1.12) for nitrification. Similar Θ values were also estimated for 17-β estradiol (E2) transformation by activated sludge, ranging from 1.03 to 1.09 for different biomass concentrations (Li et al., 2005). Wick et al. (2009) considered temperature-dependent biotransformation for successful prediction of season-dependent pharmaceutical and illicit drugs removal in WWTPs. The correction factor, Θ, for organic micropollutants such as pharmaceuticals was estimated in the range of 1.03–1.09 (Joss et al., 2006). Overall, previous and current findings demonstrate that temperature can have considerable impact on transformation, the extent of which is compound-dependent.

3.2. Influence of temperature on back-calculation of drug use

As expected, higher temperature resulted in higher in-sewer removal, with 40% (6-MAM) to almost 4-fold (EME) increase of removal efficiency from medium to high temperature. Consistently, 6-MAM and EME have lowest and highest Θ values (Table 2).

These results indicate that accounting for in-sewer transformation is important especially at elevated temperatures (above 15°C). Consequently, the temperature dependency of *k* should be accounted for explicitly in steady-state and dynamic model simulations. From this stand point, the Arrhenius equation can be included in existing modeling frameworks for removal of drug biomarkers in wastewater such as WATS—ASM-X (Ramin et al., 2016). We note that, in this study, the estimation of in-sewer removal was performed based on individual biomarkers, and the transformation of biomarkers into/from other biomarkers was neglected. It is common practice to back-calculate the consumption of

COC based on the concentration of its metabolite benzoylecgonine (BE) and COC itself. It has been found that BE, beside formation, also under go transformation (McCall et al., 2016; Ramin et al., 2016), although some studies reported negligible in-sewer BE transformation (Bisceglia and Lippa, 2014b; Thai et al., 2014). Further discussion on back-calculation of illicit drug consumption interested readers are referred to available literature (Castiglioni et al., 2013; Khan and Nicell, 2011).

It is evident that further research is crucial for obtaining new evidence on drug stability at different temperatures, especially for new psychoactive substances. This is generally relevant for other types of biomarkers beyond illicit drugs which wastewater-based epidemiology has gained interests (Gracia-Lor et al., 2017). We encourage authors to report conditions at which stability tests were performed, similarly to Table 1. This would allow for better comparison and consistency evaluation among different studies.

4. Conclusions

This study represents a first attempt to describe temperature-dependent transformation (abiotic and biotic) of five illicit drug biomarkers (COC, EME, CE, NorCOC, 6-MAM) in untreated wastewater under aerobic conditions. Following conclusions are made:

- Although affected by the considerable variability of measured transformation kinetics, the Arrhenius equation could capture trends of increasing transformation rates with increasing temperature within the applicability domain (from 2–9ºC to 30–31ºC).
- Arrhenius-based equations were estimated for each biomarker and used for removal predictions during transport in ideal sewers. Up to almost 4-fold removal efficiency was observed when temperature was changed from 15 ºC to 25°C.

- These findings have considerable implications for back-calculation of drug consumption based on the analysis of untreated wastewater influents, especially for multi-catchment studies covering wide geographical areas. Further research should extend the investigation of temperature effects to (i) a larger number biomarkers; (ii) anaerobic conditions; and (iii) sewer biofilms.

Acknowledgments

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References

Baker, D.R., Kasprzyk-Hordern, B., 2011. Critical evaluation of methodology commonly used in sample collection, storage and preparation for the analysis of pharmaceuticals and illicit drugs in surface water and wastewater by solid phase extraction and liquid chromatography-mass spectrometry. J. Chromatogr. A 1218, 8036–59. doi:10.1016/j.chroma.2011.09.012

Bisceglia, K.J., Lippa, K. a, 2014a. Stability of cocaine and its metabolites in municipal wastewater - the case for using metabolite consolidation to monitor cocaine utilization. Environ. Sci. Pollut. Res. 21, 4453–4460. doi:10.1007/s11356-013-2403-5

Bisceglia, K.J., Lippa, K. a., 2014b. Stability of cocaine and its metabolites in municipal wastewater - the case for using metabolite consolidation to monitor cocaine utilization. Environ. Sci. Pollut. Res. 21, 4453–4460. doi:10.1007/s11356-013-2403-5

Carballa, M., Omil, F., Ternes, T., Lema, J.M., 2007. Fate of pharmaceutical and personal care products (PPCPs) during anaerobic digestion of sewage sludge. Water Res. 41, 2139–2150. doi:10.1016/j.watres.2007.02.012

Castiglioni, S., Bijlsma, L., Covaci, A., Emke, E., Hernández, F., Reid, M., Ort, C., Thomas, K. V., Van Nuijs, A.L.N., De Voogt, P., Zuccato, E., 2013. Evaluation of uncertainties associated with the determination of community drug use through the measurement of sewage drug biomarkers. Environ. Sci. Technol. 47, 1452–1460. doi:10.1021/es302722f

Chen, C., Kostakis, C., Irvine, R.J., Felgate, P.D., White, J.M., 2013. Evaluation of pre-analysis loss of dependent drugs in wastewater: Stability and binding assessments. Drug Test. Anal. 5, 716–721. doi:10.1002/dta.1428

Devault, D.A., Lévi, Y., Karolak, S., 2017. Applying sewage epidemiology approach to estimate illicit

231 drug consumption in a tropical context: Bias related to sewage temperature and pH. Sci. Total
232 Environ. 584-585, 252–258. doi:10.1016/j.scitotenv.2017.01.114
233 Frutiger, J., Marcarie, C., Abildskov, J., Sin, G., 2016. A Comprehensive Methodology for
234 Development, Parameter Estimation, and Uncertainty Analysis of Group Contribution Based
235 Property Models-An Application to the Heat of Combustion. J. Chem. Eng. Data 61, 602–613.
236 doi:10.1021/acs.jced.5b00750
237 Gracia-Lor, E., Castiglioni, S., Bade, R., Been, F., Castrignanò, E., Covaci, A., González-Mariño, I.,
238 Hapeshi, E., Kasprzyk-Hordern, B., Kinyua, J., Lai, F.Y., Letzel, T., Lopardo, L., Meyer, M.R.,
239 O'Brien, J., Ramin, P., Rousis, N.I., Rydevik, A., Ryu, Y., Santos, M.M., Senta, I., Thomaidis,
240 N.S., Veloutsou, S., Yang, Z., Zuccato, E., Bijlsma, L., 2017. Measuring biomarkers in
241 wastewater as a new source of epidemiological information: Current state and future perspectives.
242 Environ. Int. 99, 131–150. doi:10.1016/j.envint.2016.12.016
243 Henze, M., Gujer, W., Mino, T., Loosdrecht, M. van, 2000. Activated Sludge Models ASM1, ASM2,
244 ASM2d AND ASM3, cientific and Technical Report No 9, IWA Publishing. London.
245 Hvitved-Jacobsen, T., Vollertsen, J., Nielsen, A.H., 2013. Sewer Processes: Microbial and Chemical
246 Process Engineering of Sewer Networks, second. ed. CRC Press.
247 Joss, A., Carballa, M., Kreuzinger, N., Siegrist, H., Zabczynski, S., 2006. Human pharmaceuticals,
248 hormones and fragrances: The challenge of micropollutants in urban water management, Science
249 of The Total Environment. IWA Publishing, London. doi:10.1016/j.scitotenv.2006.10.031
250 Khan, U., Nicell, J.A., 2011. Refined sewer epidemiology mass balances and their application to
251 heroin, cocaine and ecstasy. Environ. Int. 37, 1236–1252. doi:10.1016/j.envint.2011.05.009
252 Li, F., Yuasa, A., Obara, A., Mathews, A.P., 2005. Aerobic batch degradation of 17-beta estradiol (E2)
253 by activated sludge: Effects of spiking E2 concentrations, MLVSS and temperatures. Water Res.

39, 2065–2075. doi:10.1016/j.watres.2005.02.009

Li, J., Gao, J., Thai, P.K., Sun, X., Mueller, J.F., Yuan, Z., Jiang, G., 2018. Stability of Illicit Drugs as Biomarkers in Sewers: From Lab to Reality. Environ. Sci. Technol. acs.est.7b05109. doi:10.1021/acs.est.7b05109

Mardal, M., Kinyua, J., Ramin, P., Miserez, B., van Nuijs, A.L.N., Covaci, A., Meyer, M.R., 2016. Screening for illicit drugs in pooled human urine and urinated soil samples and studies on the stability of urinary excretion products of cocaine, MDMA, and MDEA in wastewater by hyphenated mass spectrometry techniques. Drug Test. Anal. doi:10.1002/dta.1957

McCall, A.K., Scheidegger, A., Madry, M.M., Steuer, A.E., Weissbrodt, D.G., Vanrolleghem, P.A., Kraemer, T., Morgenroth, E., Ort, C., 2016. Influence of different sewer biofilms on transformation rates of drugs. Environ. Sci. Technol. 50, 13351–13360. doi:10.1021/acs.est.6b04200

Ort, C., van Nuijs, A.L.N., Berset, J.D., Bijlsma, L., Castiglioni, S., Covaci, A., de Voogt, P., Emke, E., Fatta-Kassinos, D., Griffiths, P., Hernández, F., González-Mariño, I., Grabic, R., Kasprzyk-Hordern, B., Mastroianni, N., Meierjohann, A., Nefau, T., Östman, M., Pico, Y., Racamonde, I., Reid, M., Slobodnik, J., Terzic, S., Thomaidis, N., Thomas, K. V., 2014. Spatial differences and temporal changes in illicit drug use in Europe quantified by wastewater analysis. Addiction 109, 1338–1352. doi:10.1111/add.12570

Plósz, B.G., Reid, M.J., Borup, M., Langford, K.H., Thomas, K. V., 2013. Biotransformation kinetics and sorption of cocaine and its metabolites and the factors influencing their estimation in wastewater. Water Res. 47, 2129–2140. doi:10.1016/j.watres.2012.12.034

Ramin, P., Brock, A.L., Polesel, F., Causanilles, A., Emke, E., de Voogt, P., Plósz, B.G., 2016. Transformation and sorption of illicit drug biomarkers in sewer systems : understanding the role of

suspended solids in raw wastewater. Environ. Sci. Technol. 50, 13397–13408.

Schwaab, M., Lemos, L.P., Pinto, J.C., 2007. Optimum reference temperature for reparameterization of the Arrhenius equation. Part 1: Problems involving one kinetic constant. Chem. Eng. Sci. 62, 2750–2764. doi:10.1016/j.ces.2008.03.010

Senta, I., Krizman, I., Ahel, M., Terzic, S., 2014. Assessment of stability of drug biomarkers in municipal wastewater as a factor influencing the estimation of drug consumption using sewage epidemiology. Sci. Total Environ. 487, 659–665. doi:10.1016/j.scitotenv.2013.12.054

Thai, P.K., Jiang, G., Gernjak, W., Yuan, Z., Lai, F.Y., Mueller, J.F., 2014. Effects of sewer conditions on the degradation of selected illicit drug residues in wastewater. Water Res. 48, 538–547. doi:10.1016/j.watres.2013.10.019

van Nuijs, A.L.N., Abdellati, K., Bervoets, L., Blust, R., Jorens, P.G., Neels, H., Covaci, A., 2012. The stability of illicit drugs and metabolites in wastewater, an important issue for sewage epidemiology? J. Hazard. Mater. 239-240, 19–23. doi:10.1016/j.jhazmat.2012.04.030

Wick, A., Fink, G., Joss, A., Siegrist, H., Ternes, T.A., 2009. Fate of beta blockers and psycho-active drugs in conventional wastewater treatment. Water Res. 43, 1060–1074. doi:10.1016/j.watres.2008.11.031

Figures

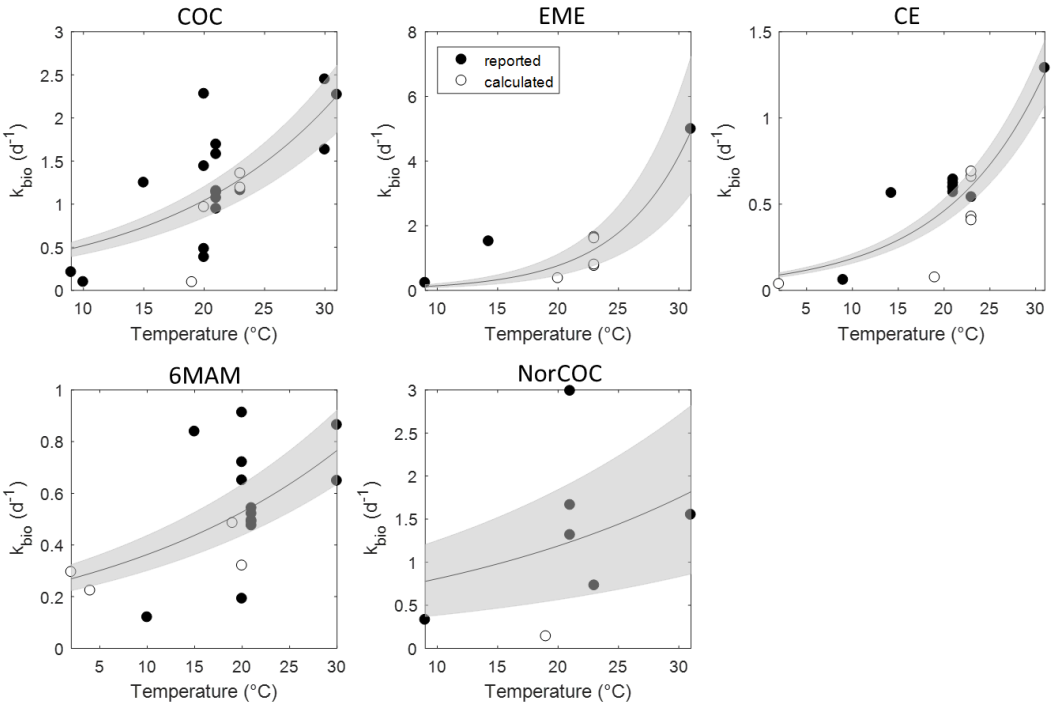

Figure 1. Arrhenius equation fits for degradation rates *k* (d^{-1}) as a function of temperature (ºC). These are based on the reported (full circles) and the estimated (empty circles) empirical values from literature. Lines are the best prediction and the shaded band is the 95% confidence interval of the prediction.

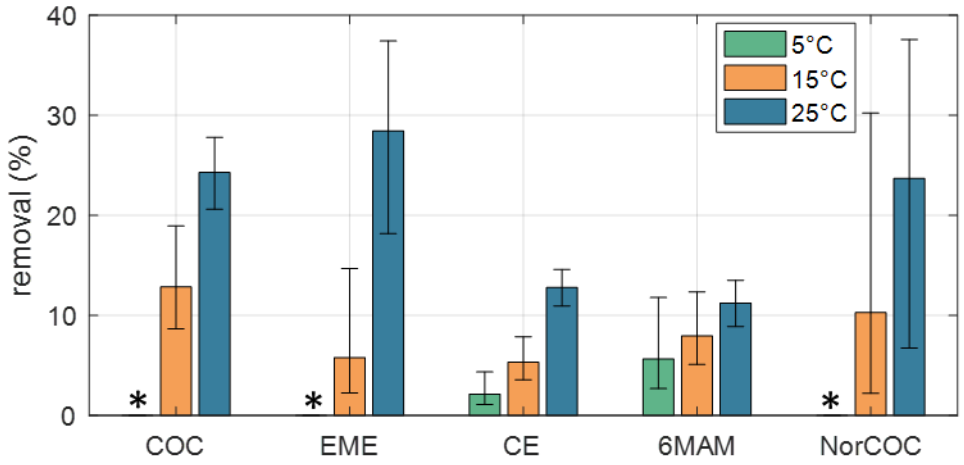

Figure 2. Estimated removal efficiencies from excretion point to WWTP influent (in-sewer residence time = 4.5 h) for selected drug biomarkers, based on the identified Arrhenius regressions. Error bars represent 95% confidence interval following Monte Carlo simulation. Asterics (*) indicates that the temperature is out of applicability range.

305 **Table 1.** Overview of selected biomarker stability studies from published literature.

	Reference	Chemical	Data source for extraction of *k*	Temp. (ºC)	pH	DO (mg L^{-1})	Duration of experiment (h)	No. of samples taken	C_0 (µg L^{-1})	TSS (g L^{-1})
1	(Baker and Kasprzyk-Hordern, 2011)	COC, CE, 6MAM, NorCOC	Table	2, 19	7.4	-	72	4	1.0	-
2	(van Nuijs et al., 2012)	COC, EME, 6MAM	Graph	20	7.5	-	26	13	0.06–0.60	-
3	(Chen et al., 2013)	6MAM	Graph	4	7.4	-	336	6	>0.1	-
4	(Bisceglia and Lippa, 2014a)	COC, EME, CE, NorCOC	Values reported	9, 23, 31	7.4	-	26	16	1.5–3.0	-
5	(Senta et al., 2014)	COC, 6MAM	Graph, values reported	20	7.5	-	72	7	0.2	-
6	(Thai et al., 2014)	COC, 6MAM	Values reported	20	7.5	-	12	9	10	-
7	(Mardal et al., 2016)	COC, EME, CE	Graph, Table	23	7-8	-	24	9	0.5-100	-
8	(Ramin et al., 2016)	COC, EME, CE, 6MAM	Values reported	14	8.6–8.8	10	48	9	10	0.32 ± 0.04
9	(McCall et al., 2016)	COC, CE, 6MAM, NorCOC	Values reported	21	8.0–8.9	5–8	24	11	2.0–3.0	0.14–0.29
10	(Devault et al., 2017)	COC, 6MAM	Values reported	20, 30	6.6, 7.6	-	24	7	1.0–3.0	-

[1]Used silanized amber glass bottles stored in the dark.
[2]Stability test performed in silanized glass flasks which were hand-shaken app. 10 times per hour.
[3]Bottles at 20ºC were placed under fume cupboard uncapped and gently stirred 3 times per day (distilled water was used to compensate for evaporation). Bottle at 4ºC was stored with cap on.
[4]Used Erlenmeyer flask equipped with foam stopper to allow air transfer. Reactor was shaken at 180 rpm in the dark.
[5]Glass bottles were capped with cotton plugs and placed in a thermostated cabinet.
[6]Used gravity sewer reactor with continuous mixing with magnetic stirrer (250 rpm) to enhance surface aeration.
[7]Urinary samples collected at a music festival was dilutted with wastewater and incubated in a temperature water bath
[8]Transformation study was performed in a covered jacketed reactor equipped with an agitator and oxygen diffuser.
[9]Transformaiton study was conducted in Erlenmeyer flask on a shaker table in the dark. Autoclaved wastewater was chosen to represent abiotic transformation.
[10]Glass bottles were placed in the dark and aerobic conditinos was maintained by shaking with a magnetic stir bar.

306 **Table 2.** Estimated k_{T25} (d^{-1}) and θ and their correlation for the selected drugs. Parameters are estimated
307 as the best fitted value together with 95% confidence interval. The predictions are valid in the reported
308 temperature range.

	k_{T25} (d^{-1})	Θ	Correlation (k_{T25} and Θ)	Temperature range (ºC)
COC	1.48 (1.23, 1.75)	1.07 (1.04, 1.11)	0.06	9–31
EME	1.78 (1.03, 2.54)	1.18 (1.09, 1.28)	-0.75	9–31
CE	0.73 (0.61, 0.85)	1.10 (1.06, 1.13)	0.07	2–31
6MAM	0.64 (0.49, 0.78)	1.04 (1.00, 1.07)	0.49	2–30
NorCOC	1.44 (0.32, 2.57)	1.04 (0.90, 1.18)	0.29	9–31

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Table 1. Overview of selected biomarker stability studies from published literature.

	Reference	Chemical	Data source for extraction of *k*	Temp. (°C)	pH	DO (mg L^{-1})	Duration of experiment (h)	No. of samples taken	C_0 (µg L^{-1})	TSS (g L^{-1})
1	(Baker and Kasprzyk-Hordern, 2011)	COC, CE, 6MAM, NorCOC	Table	2, 19	7.4	-	72	4	1.0	-
2	(van Nuijs et al., 2012)	COC, EME, 6MAM	Graph	20	7.5	-	26	13	0.06–0.60	-
3	(Chen et al., 2013)	6MAM	Graph	4	7.4	-	336	6	>0.1	-
4	(Bisceglia and Lippa, 2014a)	COC, EME, CE, NorCOC	Values reported	9, 23, 31	7.4	-	26	16	1.5–3.0	-
5	(Senta et al., 2014)	COC, 6MAM	Graph, values reported	20	7.5	-	72	7	0.2	-
6	(Thai et al., 2014)	COC, 6MAM	Values reported	20	7.5	-	12	9	10	-
7	(Mardal et al., 2016)	COC, EME, CE	Graph, Table	23	7-8	-	24	9	0.5-100	-
8	(Ramin et al., 2016)	COC, EME, CE, 6MAM	Values reported	14	8.6–8.8	10	48	9	10	0.32 ± 0.04
9	(McCall et al., 2016)	COC, CE, 6MAM, NorCOC	Values reported	21	8.0–8.9	5–8	24	11	2.0–3.0	0.14–0.29
10	(Devault et al., 2017)	COC, 6MAM	Values reported	20, 30	6.6, 7.6	-	24	7	1.0–3.0	-

[1]Used silanized amber glass bottles stored in the dark.
[2]Stability test performed in silanized glass flasks which were hand-shaken app. 10 times per hour.
[3]Bottles at 20°C were placed under fume cupboard uncapped and gently stirred 3 times per day (distilled water was used to compensate for evaporation). Bottle at 4°C was stored with cap on.
[4]Used Erlenmeyer flask equipped with foam stopper to allow air transfer. Reactor was shaken at 180 rpm in the dark.
[5]Glass bottles were capped with cotton plugs and placed in a thermostated cabinet.
[6]Used gravity sewer reactor with continuous mixing with magnetic stirrer (250 rpm) to enhance surface aeration.
[7]Urinary samples collected at a music festival was dilutted with wastewater and incubated in a temperature water bath
[8]Transformation study was performed in a covered jacketed reactor equipped with an agitator and oxygen diffuser.
[9]Transformaiton study was conducted in Erlenmeyer flask on a shaker table in the dark. Autoclaved wastewater was chosen to represent abiotic transformation.
[10]Glass bottles were placed in the dark and aerobic conditinos was maintained by shaking with a magnetic stir bar.

Table 2. Estimated k_{T25} (d^{-1}) and θ and their correlation for the selected drugs. Parameters are estimated as the best fitted value together with 95% confidence interval. The predictions are valid in the reported temperature range.

	k_{T25} (d^{-1})	Θ	Correlation (k_{T25} and Θ)	Temperature range (ºC)
COC	1.48 (1.23, 1.75)	1.07 (1.04, 1.11)	0.06	9–31
EME	1.78 (1.03, 2.54)	1.18 (1.09, 1.28)	-0.75	9–31
CE	0.73 (0.61, 0.85)	1.10 (1.06, 1.13)	0.07	2–31
6MAM	0.64 (0.49, 0.78)	1.04 (1.00, 1.07)	0.49	2–30
NorCOC	1.44 (0.32, 2.57)	1.04 (0.90, 1.18)	0.29	9–31

Figure
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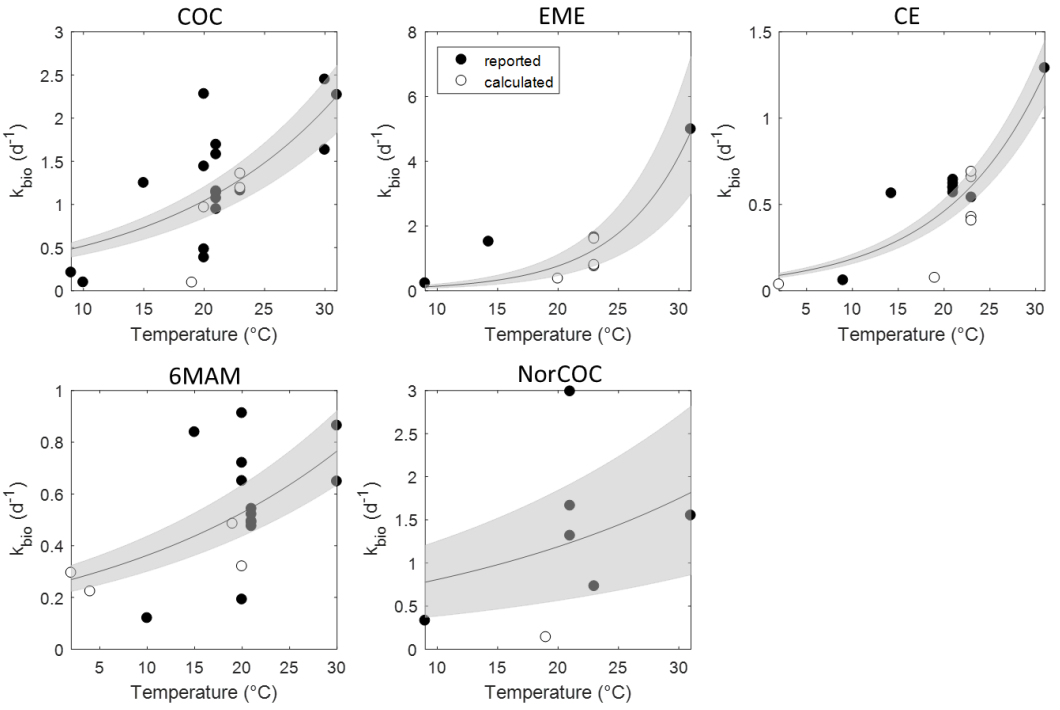

Figure 1. Arrhenius equation fits for degradation rates *k* (d^{-1}) as a function of temperature (ºC). These are based on the reported (full circles) and the estimated (empty circles) empirical values from literature. Lines are the best prediction and the shaded band is the 95% confidence interval of the prediction.

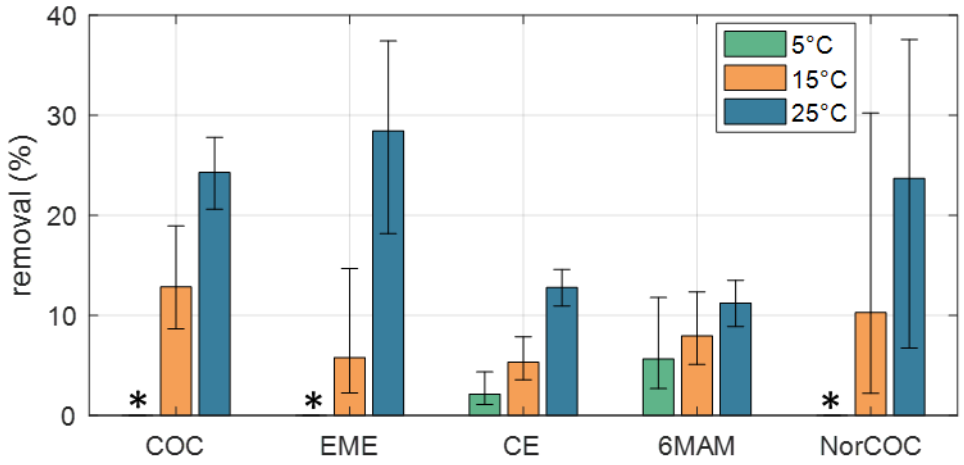

Figure 2. Estimated removal efficiencies from excretion point to WWTP influent (in-sewer residence time = 4.5 h) for selected drug biomarkers, based on the identified Arrhenius regressions. Error bars represent 95% confidence interval following Monte Carlo simulation. Asterics (*) indicates that the temperature is out of applicability range.